

Timing in Free-Living Rufous Hummingbirds, *Selasphorus rufus*

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Summary

Animals organize their lives around circannual and circadian rhythms, but little is known of their use of much shorter intervals [1]. In the laboratory, some animals can learn the specific duration (seconds or minutes) between periods of food access [2]. It has been supposed that wild nectarivores, such as hummingbirds, might also learn short time intervals so as to avoid re-visiting emptied flowers until the nectar has been replenished [3]. We provided free-living, territorial rufous hummingbirds each with eight artificial flowers containing sucrose solution. Four flowers were refilled 10 min after the bird emptied them, and the other four were refilled 20 min after being emptied. Throughout the day, birds revisited the 10 min flowers significantly sooner than they revisited the 20 min flowers, and return visits to the flowers matched their refill schedules. Hummingbirds remembered the locations and timing of eight rewards, updating this information throughout the day. Not only is this the first time that this degree of timing ability has been shown in wild animals, but these hummingbirds also exhibit two of the fundamental aspects of episodic-like memory (where and when [4, 5]), the kind of memory for specific events often thought to be exclusive to humans [6, 7].

Results and Discussion

Many important events in animals' lives, from the availability of food to the arrival of predators, occur predictably in time, and many animals are able to gauge the intervals between these events [e.g., 2, 8]. Timing abilities range in scale from animals varying activity levels according to events (such as the predictable onset of dawn and dusk) to anticipating the availability of food over shorter intervals. Other events, however, have

a far less predictable periodicity and, in the case of nectarivores feeding on renewing food sources, the periodicity is, to a large extent, dependent on their own actions. Because emptied flowers might take several hours to replenish, hummingbirds could save time and energy by avoiding them. To do this, the bird should remember not only where the flower was, but also when it was last visited. Territorial rufous hummingbirds avoid recently emptied flowers by remembering where the flowers were [9, 10], and traplining hummingbirds show some evidence of being able to track temporal changes [11, 12].

This experiment was designed to determine whether free-living rufous hummingbirds could remember when they had visited a number of artificial flowers (see Figure 1), which were then refilled at intervals of either 10 or 20 min after being emptied. After a number of days, we compared the timing of the birds' visits made to both 10 and 20 min flowers. Visits were defined as probes made by the bird to individual flowers within a foraging bout. The intervals between visits to each flower were calculated for each experimental session. Frequency distributions of all "postreinforcement pauses" (PRPs), the intervals between a bird's current visit to a flower and its last rewarded visit to that flower, were then constructed (Figure 2). Because interbout intervals were determined by the birds, and tended to be less than 10 min (Bird 1: 8 min; Bird 2: 10 min; Bird 3: 6 min), birds had frequent opportunities to visit all the flowers and yet decline visits to flowers that had been emptied during the recent foraging bouts.

The patterns of visitation to the array support three primary inferences. The first is that birds learned that the refill rates differed among the flowers. Birds revisited the 10 min flowers significantly sooner than they visited 20 min flowers (Mann Whitney U tests, adjusted for ties: Bird 1, $n = 2271$; Bird 2, $n = 2504$; Bird 3, $n = 3277$; $p < 0.0001$ in all three cases). Critically, the PRPs for 10 min flowers were significantly shorter than the value 20 (Wilcoxon Signed Rank Test: $p < 0.001$ in all three cases), indicating that the birds avoided the 20 min flowers when they returned to the array sooner than 20 min after the previous bout. Given that all of the flowers differed in their color and pattern and they were not arranged by refill rate, it seems that the birds do not use relative familiarity of the array to determine which flowers should be visited [13, 14].

The second inference that can be drawn from these data is that the hummingbirds learned that the specific refill times were 10 and 20 min. Whereas many studies indicate that animals anticipate rewards, and indeed many of the individual hummingbird PRPs were shorter than the refill rate, the birds' central tendency was to match or exceed the refill rate. Not one of the median values was shorter than the refill rate (Wilcoxon 1-tailed, 1-sample tests, 10 minutes: Bird 1, median = 14 min, $n = 1231$; Bird 2, median = 13 min, $n = 1361$; Bird 3, median = 12 min, $n = 1933$; for all, $p < 0.001$; 20 minutes: Bird 1,

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Figure 1. A Male Rufous Hummingbird Feeding from One of the Experimental Flowers. The red ink mark on his chest was for individual identification.

median = 20 min, $n = 1050$, $p = 0.51$; Bird 2, median = 21 min, $n = 1143$, $p = 0.001$; Bird 3, median = 20 min, $n = 1344$, $p = 0.07$). An alternative measure of central

tendency is the mode. Modal PRPs matched refill rates very closely (10/20 min flowers: Bird 1: 11/21 min; Bird 2: 10/20 min; Bird 3: 10/20 min; Figure 2).

Finally, the hummingbirds distinguished between 10 min and 20 min flowers, in general, and they also seem to have remembered specifically which flowers they had emptied recently. Such abilities have been shown only in laboratory experiments in which animals receive extensive training. Not only were the hummingbirds able to learn the refill schedules, as shown in other animals in the laboratory, but more remarkably, the hummingbirds were also able to update this information across numerous foraging bouts throughout the day. Such updating abilities have rarely been demonstrated in any animal.

It is to be expected that animals increase their responding rate in concert with reinforcement rate when faced with concurrent choices offering different rates of reward (the matching law [15–17]). All birds visited 10 min flowers significantly more frequently than 20 min flowers (Wilcoxon Signed Rank Test: all $p < 0.004$). Ten minute flowers were not, however, visited twice as often as 20 min flowers (ratio of visits to 10:20 min flowers: Bird 1: 1.08, Bird 2: 1.20, Bird 3: 1.43; all tested against $H_0 = 1$, all $p < 0.004$; tested against $H_0 = 2$, all $p < 0.001$). Such a deviation from strict matching is, however, also common to other studies [18, 19].

Across days, birds varied in how well their visits matched the refill schedule. From the first session onward for all three birds, their visits to the 10 min flowers always matched, or exceeded, 10 min. In contrast, revisiting of the 20 min flowers took a little over two sessions before the PRPs were consistently around 20 min or longer (Figure 3). Given that they visited these flowers less frequently than the 10 min flowers, it is perhaps not surprising that it took the birds longer to learn the refill schedule. This is also consistent with the finding that longer intervals are harder to time than shorter ones (see e.g., [20]), as a result of increased uncertainty in estimating longer intervals [21]. However, this should also result in the standard deviation of timing an interval being proportional to the magnitude of that interval (see e.g., [16]), which was not the case for any of the birds: Although the coefficients of variation did not differ

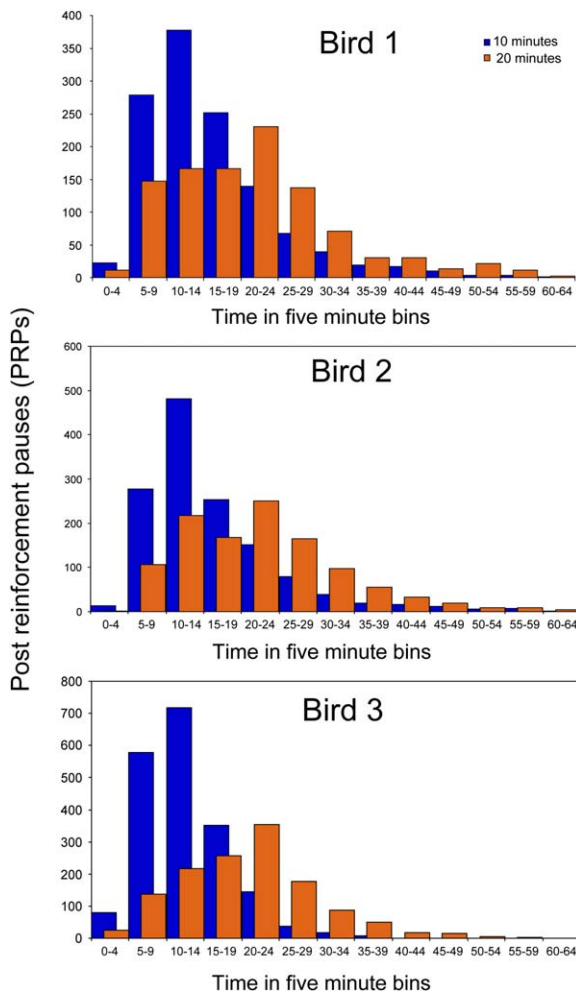


Figure 2. Frequency Distributions of the Number of Postreinforcement Pauses for Five Minute Bins for Birds 1, 2, and 3. 10 min data are colored blue and 20 min data are colored orange.

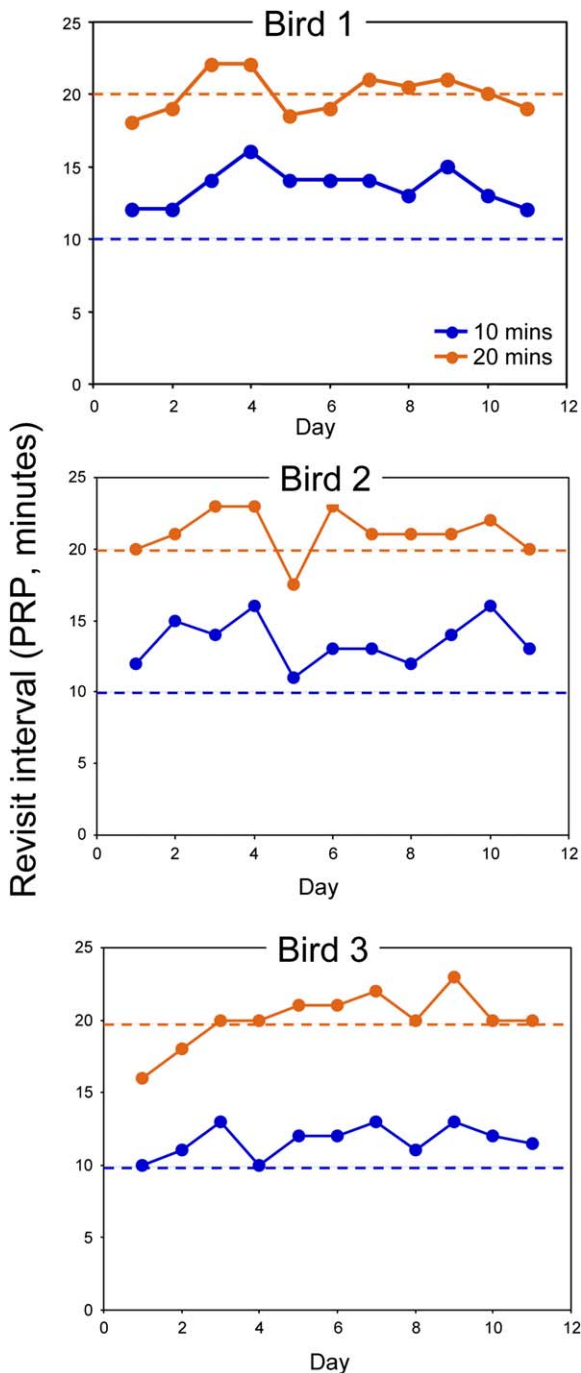


Figure 3. Median Postreinforcement Pauses for Birds 1, 2, and 3 for Each Day

The blue circles represent 10 min data, the dashed blue line denotes 10 min, the orange circles represent 20 min data, and the dashed orange line denotes 20 min.

between the two refill rates (paired *t* test: $t = 1.37$, $p = 0.31$, $df = 2$), the standard deviations (SDs) for 10 min were significantly different from those for 20 min (mean $SD_{10} = 8.14$, mean $SD_{20} = 11.18$; $t = 6.69$, $p = 0.02$, $df = 2$). All the birds learned the two intervals more readily, if not quite as accurately, as animals trained in the lab, and the birds timed the 20 min refill flowers better than would have been expected. It must also be

remembered that these birds were both “in control” of the experiment and constantly occupied with other important behaviors throughout the experiment, such as displaying to females and guarding their territory against intruding conspecific males. Their timing ability seems all the more impressive given these competing calls on their attention and time.

To our knowledge, this is the first demonstration that free-living animals remember both the locations of multiple rewards and when they visited each of those locations. Additionally, in order to continue doing this accurately through the day and between days, the birds had to be able to update their memories continually for each flower. Because natural flowers differ in nectar level and concentration, and hummingbirds are able to remember each of these [22], they may well be able to remember all three components of a flower (where, when, and what). Territorial hummingbirds would, then, like food-storing birds, appear to make use of episodic-like memories in their everyday lives [23, 24]. Although there are both rat and primate episodic-like-memory models in which the animals remember the what and where of an item or event [25–28], it is the temporal component of a memory that has been difficult to demonstrate ([25, 29], but see [30]). Keeping track of the timing of past events is, however, a constant part of daily life for a foraging, territorial hummingbird. The timing abilities of these birds, thus, provide a useful contribution to the current debate on episodic-like-memory processes and whether or not they exist in animals other than humans.

Experimental Procedures

The subjects used in this experiment were three wild, male rufous hummingbirds. The experiment was run from 0800–2000 hr Mountain Standard Time in June and July 2000 in a valley in the eastern Rocky Mountains, Alberta, Canada. Rufous hummingbirds migrate to this valley to breed, and males set up territories centered around artificial feeders.

It took 1–2 hr to train each bird to feed from artificial flowers, containing sucrose, placed in his territory. A flower consisted of a white cardboard disc (6 cm diameter), mounted horizontally on a wooden stake (60 cm high). At the center of the flower was a blue syringe tip (capable of holding 120 μ l) in a small cork mounted on top of the stake. When the bird was consistently feeding from these flowers, the experiment began.

At the start of a day’s testing, the bird’s feeder was removed and an array of eight flowers was placed in his territory, in the same place for each session. The array consisted of eight flowers in a 3,2,3 configuration with a nearest-neighbor distance of 60 cm. The flowers were the same as in training except that each was of a different color (neon green, orange, blue, brown, purple, yellow, green, red). Each flower contained 20 μ l (concentration 20%) sucrose solution. Once the bird had visited the array for the first time, two different fixed-interval flower refill schedules were used. Four flowers were designated as 10 min flowers, i.e., each was refilled 10 min after the bird had emptied that flower, the other four as 20 min flowers, i.e., refilled 20 min after being emptied. The locations of 10 and 20 min flowers within the array were determined in a pseudorandom fashion and remained the same throughout the experiment. The male was free to visit flowers throughout the day. At the end of each session, the array was removed and the bird’s feeder replaced. Small markers were placed in the ground to ensure that in subsequent sessions the array was put back in the same place. The time of the bird’s visits to the array and the flowers visited in each bout were recorded throughout each session. The total number of bouts varied across birds (Bird 1, 536 bouts across 13 sessions; Bird 2, 495 bouts across 11 sessions; and Bird 3, 695 bouts across 12 sessions).

To ensure that the results we observed above were not an artifact of the birds' repeatedly using the same visiting strategy, one that happened to interact with our experimental refill rate, which could then lead to a bias in the temporal distribution of the data, we also repeated all of the analyses by using only the data from the first visit birds made to a flower following their last rewarded visit (the first PRP). However, although there were many fewer data, of the outcomes reported above, the only difference seen between the two data measures is that the birds returned to the 20 min flowers sooner than 20 min. Nonetheless, they still returned to 10 min flowers significantly sooner than to the 20 min flowers.

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References

1. Carr, J.A.R., and Wilkie, D.M. (1997). Ordinal, phase, and interval timing. In *Time and Behaviour: Psychological and Neurobehavioural Analyses*, C.M. Bradshaw and E. Szabadi, eds. (New York: Elsevier), pp. 265–327.
2. Gallistel, C.R. (1990). *The Organization of Learning* (Cambridge, MA: MIT Press).
3. Healy, S.D., and Hurly, T.A. (2004). Spatial learning and memory in birds. *Brain Behav. Evol.* *63*, 211–220.
4. Clayton, N.S., and Dickinson, A. (1998). Episodic-like memory during cache recovery by scrub jays. *Nature* *395*, 272–274.
5. Morris, R.G.M. (2001). Episodic-like memory in animals: Psychological criteria, neural mechanisms and the value of episodic-like tasks to investigate animal models of neurodegenerative disease. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* *356*, 1453–1465.
6. Suddendorf, T., and Busby, J. (2003). Mental time travel in animals? *Trends Cogn. Sci.* *7*, 391–396.
7. Tulving, E. (1983). *Elements of Episodic Memory* (Oxford: Clarendon Press).
8. Gibbon, J., Malapani, C., Dale, C.L., and Gallistel, C.R. (1997). Toward a neurobiology of temporal cognition: Advances and challenges. *Curr. Opin. Neurobiol.* *7*, 170–184.
9. Healy, S.D., and Hurly, T.A. (1995). Spatial memory in rufous hummingbirds (*Selasphorus rufus*): A field test. *Anim. Learn. Behav.* *23*, 63–68.
10. Henderson, J., Hurly, T.A., and Healy, S.D. (2001). Rufous hummingbirds' memory for flower location. *Anim. Behav.* *61*, 981–986.
11. Garrison, J.S.E., and Gass, C.L. (1999). Response of a traplining hummingbird to changes in nectar availability. *Behav. Ecol.* *10*, 714–725.
12. Gill, F.B. (1988). Trapline foraging by hermit hummingbirds: Competition for an undefended renewable resource. *Ecology* *69*, 1933–1942.
13. Jacoby, L.L. (1991). A process dissociation framework: Separating automatic from intentional uses of memory. *J. Mem. Lang.* *30*, 513–541.
14. de Kort, S.R., Dickinson, A., and Clayton, N.S. (2005). Retrospective cognition by food-caching western scrub-jays. *Learn. Motiv.* *36*, 159–176.
15. Miller, H.L., and Loveland, D.H. (1974). Matching when the number of response alternatives is large. *Anim. Learn. Behav.* *2*, 106–110.
16. Church, R.M., and Gibbon, J. (1982). Temporal generalization. *J. Exp. Psychol. Anim. Behav. Processes.* *8*, 165–186.
17. Rider, D.P. (1987). Temporal control of responding under mixed fixed-interval schedules of reinforcement. *Psychol. Rec.* *37*, 409–422.
18. Bacotti, A.V. (1977). Matching under concurrent fixed-ratio variable-interval schedules of food presentation. *J. Exp. Anal. Behav.* *25*, 171–182.
19. Davison, M., and McCarthy, D. (1988). *The Matching Law: A Research Review* (Hillsdale, New Jersey: Lawrence Erlbaum Associates).
20. Brodbeck, D.R., Hampton, R.R., and Cheng, K. (1998). Timing behaviour of black-capped chickadees (*Parus atricapillus*). *Behav. Processes* *44*, 183–195.
21. Malapani, C., and Fairhurst, S. (2002). Scalar timing in animals and humans. *Learn. Motiv.* *33*, 156–176.
22. Bateson, M., Healy, S.D., and Hurly, T.A. (2003). Context-dependent foraging decisions in rufous hummingbirds. *Proc. R. Soc. Lond. B. Biol. Sci.* *270*, 1271–1276.
23. Clayton, N.S., and Dickinson, A. (1999). Scrub jays (*Aphelocoma coerulescens*) remember the relative time of caching as well as the location and content of their caches. *J. Comp. Psychol.* *113*, 403–416.
24. Clayton, N.S., Yu, K.S., and Dickinson, A. (2003). Interacting cache memories: Evidence for flexible memory use by Western Scrub-Jays (*Aphelocoma californica*). *J. Exp. Psychol. Anim. Behav. Processes.* *29*, 14–22.
25. Bird, L.R., Roberts, W.A., Abroms, B., Kit, K.A., and Crupi, C. (2003). Spatial memory for food hidden by rats (*Rattus norvegicus*) on the radial maze: Studies of memory for where, what, and when. *J. Comp. Psychol.* *117*, 176–187.
26. Eacott, M.J., and Norman, G. (2004). Integrated memory for object, place and context in rats: A possible model of episodic-like memory? *J. Neurosci.* *24*, 1948–1953.
27. Schwartz, B.L., Hoffman, M.L., and Evans, S. (2005). Episodic-like memory in a gorilla: A review and new findings. *Learn. Motiv.* *36*, 226–244.
28. Menzel, C.R. (1999). Unprompted recall and reporting of hidden objects by a chimpanzee (*Pan troglodytes*) after extended delays. *J. Comp. Psychol.* *113*, 426–434.
29. Hampton, R.R., Hampstead, B.M., and Murray, E.A. (2005). Rhesus monkeys (*Macaca mulatta*) demonstrate robust memory for what and where, but not when, in an open-field test of memory. *Learn. Motiv.* *36*, 245–259.
30. Babb, S.J., and Crystal, J.D. (2005). Discrimination of what, when, and where: Implications for episodic-like memory in rats. *Learn. Motiv.* *36*, 177–189.